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Docket No.: 58884US002

Transmittal of Provisional Application

Mail Stop Provisional Patent Application
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Inventor(s): Tushar Kshirsagar, Woodbury, Minnesota

Title: HYDROXYLAMINE SUBSTITUTED IMIDAZOQUINOLINES

1. ☒ Enclosed is the above-identical new provisional application for patent under 35 USC § 111(b)(1). It includes:
   53 Pages of Text
   0 Sheets of Drawings
2. ☐ Enclosed is an executed Assignment to 3M Innovative Properties Company and a completed Assignment Recordation Cover Sheet.
3. ☐ This invention was made under a contract with an agency of the U.S. Government:
   Agency: _____
   Contract No. _____
4. ☒ Correspondence Address: Dean A. Ersfeld
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5. ☒ Please charge the $160.00 filing fee under 37 CFR § 1.16(k) to Deposit Account No. 13-3723. One copy of this sheet marked duplicate is also enclosed.
6. ☒ Please charge to Deposit Account No. 13-3723 any fees under 37 CFR §§ 1.16 and 1.17, which may be required to file and during the entire pendency of this application. This authorization includes the fee for any necessary extension of time under 37 CFR § 1.136(a). To the extent any such extension should become necessary, it is hereby requested.
7. ☒ Enclosed is a return receipt postcard.

Respectfully submitted,

12 August 2003
Date

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Filing of Papers and Fees by Express Mailing

Pursuant to 37 CFR § 1.10, this application and the documents and fees listed on this transmittal letter are being deposited on the date indicated below with the United States Postal Service "Express Mail Post Office to Addressee" service addressed to: Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450

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HYDROXYLAMINE SUBSTITUTED IMIDAZOQUINOLINES

BACKGROUND

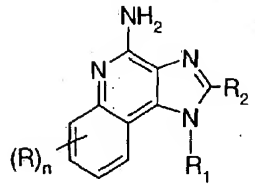
In the 1950's the 1*H*-imidazo[4,5-*c*]quinoline ring system was developed, and 1-(6-methoxy-8-quinolinyl)-2-methyl-1*H*-imidazo[4,5-*c*]quinoline was synthesized for possible use as an antimalarial agent. Subsequently, syntheses of various substituted 1*H*-imidazo[4,5-*c*] quinolines were reported. For example, 1-[2-(4-piperidyl)ethyl]-1*H*-imidazo[4,5-*c*]quinoline was synthesized as a possible anticonvulsant and cardiovascular agent. Also, several 2-oxoimidazo[4,5-c]quinolines have been reported.

Certain 1*H*-imidazo[4,5-*c*]quinolin-4-amines and 1- and 2-substituted derivatives thereof were later found to be useful as antiviral agents, bronchodilators and immunomodulators.

There continues to be interest in the imidazoquinoline ring system and there is a continuing need for compounds that have the ability to modulate the immune response, by induction of cytokine biosynthesis or other mechanisms.

SUMMARY

The present invention provides a new class of compounds that are useful in inducing cytokine biosynthesis in animals. Such compounds are of the following Formula (I):



I

wherein: R, n, R_1, and R_2 are as defined below.

The compounds of Formula I are useful as immune response modifiers due to their ability to induce cytokine biosynthesis (e.g., induces the synthesis of at least one cytokine) and otherwise modulate the immune response when administered to animals. This makes the compounds useful in the treatment of a variety of conditions such as viral diseases and tumors that are responsive to such changes in the immune response.

The invention further provides pharmaceutical compositions containing an effective amount of a compound of Formula I and methods of inducing cytokine biosynthesis in an animal, treating a viral infection and/or treating a neoplastic disease in an animal by administering an effective amount of a compound of Formula I to the animal.

In addition, methods of synthesizing compounds of Formula I and intermediates useful in the synthesis of these compounds are provided.

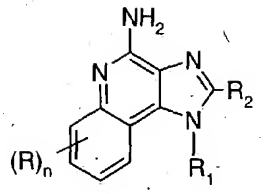
As used herein, "a," "an," "the," "at least one," and "one or more" are used interchangeably.

The terms "comprises" and variations thereof do not have a limiting meaning where these terms appear in the description and claims.

The above summary of the present invention is not intended to describe each disclosed embodiment or every implementation of the present invention. The description that follows more particularly exemplifies illustrative embodiments. In several places throughout thedescription, guidance is provided through lists of examples, which examples can be used in various combinations. In each instance, the recited list serves only as a representative group and should not be interpreted as an exclusive list.

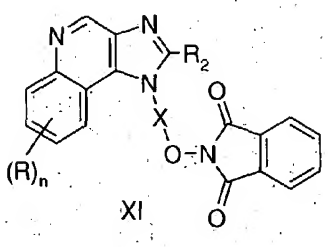
DETAILED DESCRIPTION OF ILLUSTRATIVE EMBODIMENTS OF THE INVENTION

The present invention provides compounds of the following Formula (I):

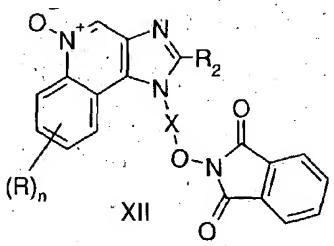


I

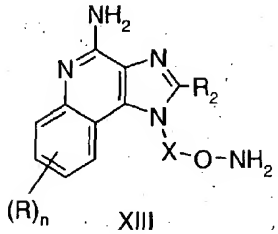
as well as intermediates of formulas (XI, XII, and XIII):



XI



XII



XIII

wherein:

R_1 is -X-Q-R_1';

X is -CH(R_4)alkylene or -CH(R_4)alkenylene;

Q is -O-NH-Z-;

Z is selected from the group consisting of:

a bond;

-C(O)-;

-C(S)-;

-$S(O)_2$-;

-$S(O)_2$-$N(R_5)$-;

-C(O)-O-;

-C(O)-$N(R_5)$-;

-C(S)-$N(R_5)$-;

-C(O)-$N(R_5)$-$S(O)_2$-;

-C(O)-$N(R_5)$-C(O)-;

-C(S)-$N(R_5)$-C(O)-; and

-C(O)-C(O)-O-;

R_1' is selected from the group consisting of:

-hydrogen;

-alkyl;

-alkenyl;

-aryl;

-alkylene-aryl;

-alkylene-heteroaryl;

-alkylene-heterocyclyl;

-heteroaryl;

-heterocyclyl; and

alkyl, alkenyl, aryl, arylalkylenyl, heteroarylalkylenyl; heterocyclylalkylenyl, heteroaryl or heterocyclyl, substituted by one or more substituents selected from the group consisting of:

-OH;

-alkyl;

-haloalkyl;

-hydroxyalkyl;

-O-alkyl;

-$S(O)_{0-2}$-alkyl;

-$S(O)_{0-2}$-aryl;

-O-haloalkyl;

-halogen;

-nitrile;

-nitro;

-aryl;

-heteroaryl;

-heterocyclyl;

-O-aryl;

-O-alkylene-aryl;

-C(O)-O-alkyl;

-$C(O)$-$N(R_5)_2$;

-$N(R_5)$-C(O)-alkyl;

-O-(CO)-alkyl; and

-C(O)-alkyl;

n is 0-4;

each R and R_2 are independently selected from the group consisting of hydrogen and non-interfering substituents;

R_4 is selected from the group consisting of hydrogen and alkyl which may be optionally interrupted by one or more -O- groups; and

each R_5 is independently selected from the group consisting of hydrogen, C_{1-10}alkyl, and C_{2-10}alkenyl;

or a pharmaceutically acceptable salt thereof.

Herein, "non-interfering" means that the immunomodulator activity of the compound is not destroyed.

For certain embodiments of Formula I, Z is -C(O)-, -$S(O)_2$-, or

$-C(O)-N(R_5)-$.

For certain embodiments of Formula I, R_1' is independently selected from the group consisting of -alkyl, -alkenyl, -aryl, and heteroaryl, each of which is optionally substituted by one or more substituents selected from the group consisting of -O-alkyl, -O-aryl, -S-alkyl, -S-aryl, halogen, -O-C(O)-alkyl, -C(O)-O-alkyl, -haloalkoxy, -haloalkyl, and -aryl. For certain other embodiments, R_1' is independently selected from the group consisting of -alkyl and -aryl, each of which is optionally substituted by one or more substituents selected from the group consisting of -O-alkyl, -O-aryl, -S-alkyl, -S-aryl, halogen, -O-C(O)-alkyl, -C(O)-O-alkyl, -haloalkoxy, -haloalkyl, and aryl.

For certain embodiments of Formulas I, XI, XII, and XIII, X is $-CH(R_4)C_{1-10}$alkylene. For other embodiments, X is propylene or butylene.

For certain embodiments of Formulas I, XI, XII, and XIII, n is 0.

For certain embodiments of Formulas I, XI, XII, and XIII, each R is independently selected from the group consisting of alkyl, alkoxy, halogen, hydroxy, and trifluoromethyl.

For certain embodiments of Formulas I, XI, XII, and XIII, R_2 is selected from the group consisting of:

-hydrogen;

-alkyl;

-alkenyl;

-aryl;

-heteroaryl;

-heterocyclyl;

-alkylene-Y-alkyl;

-alkylene-Y- alkenyl;

-alkylene-Y-aryl; and

- alkyl or alkenyl substituted by one or more substituents selected from the group consisting of:

-OH;

-halogen;

$-N(R_5)_2$;

$-C(O)-C_{1-10}$alkyl;

$-C(O)-O-C_{1-10}$alkyl;

$-N_3$;

-aryl;

-heteroaryl;

-heterocyclyl;

-C(O)-aryl; and

-C(O)-heteroaryl;

wherein:

Y is –O– or $-S(O)_{0-2}$-; and

each R_5 is independently selected from the group consisting of hydrogen, C_{1-10}alkyl, and C_{2-10}alkenyl.

For certain embodiments of Formulas I, XI, XII, and XIII, R_2 is selected from the group consisting of hydrogen, alkyl, and -alkylene-O-alkyl.

As used herein, the terms "alkyl," "alkenyl," "alkynyl," and the prefix "alk-" are inclusive of both straight chain and branched chain groups and, in the case of alkyl and alkenyl, of cyclic groups, i.e., cycloalkyl and cycloalkenyl. Unless otherwise specified, these groups contain from 1 to 20 carbon atoms, with alkenyl and alkynyl groups containing from 2 to 20 carbon atoms. The alkenyl and alkynyl groups can contain one or more double and triple bonds respectively. In some embodiments, preferred groups have a total of up to 10 carbon atoms. In other embodiments, preferred groups have a total of up to 8, up to 6, or up to 4 carbon atoms. Cyclic groups can be monocyclic or polycyclic and preferably have from 3 to 10 ring carbon atoms. Exemplary cyclic groups include cyclopropyl, cyclopentyl, cyclohexyl, cyclopropylmethyl, adamantyl, norbornane, and norbornene.

The terms "alkylene," "alkenylene," and "alkynylene" are inclusive of divalent radicals derived from the alkyl, alkenyl, and alkynyl groups described above.

The term "haloalkyl" is inclusive of groups that are substituted by one or more halogen atoms, including perfluorinated groups. This is also true of groups that include the prefix "halo-." Examples of suitable haloalkyl groups are chloromethyl, trifluoromethyl, and the like.

The term "aryl" as used herein includes carbocyclic aromatic rings or ring systems. Examples of aryl groups include phenyl, naphthyl, biphenyl, fluorenyl and indenyl. The term "heteroaryl" includes aromatic rings or ring systems that contain at least one ring heteroatom (e.g., O, S, N). Suitable heteroaryl groups include furyl, thienyl, pyridyl, quinolinyl, isoquinolinyl, indolyl, isoindolyl, triazolyl, pyrrolyl, tetrazolyl, imidazolyl, pyrazolyl, oxazolyl, thiazolyl, benzofuranyl, benzothiophenyl, carbazolyl, benzoxazolyl, pyrimidinyl, benzimidazolyl, quinoxalinyl, benzothiazolyl, naphthyridinyl, isoxazolyl, isothiazolyl, purinyl, quinazolinyl, and so on.

The term "alkylene-aryl" as used herein includes alkyl groups substituted by aromatic rings as well as cycloalkyl groups fused to aromatic rings. Exemplary groups include benzyl, phenethyl, indanyl, and tetrahydronaphthyl.

The terms "arylene" and "heteroarylene" include divalent radicals derived from the aryl and heteroaryl groups described above.

"Heterocyclyl" includes non-aromatic rings or ring systems that contain at least one ring heteroatom (e.g., O, S, N) and includes all of the fully saturated and partially unsaturated derivatives of the above mentioned heteroaryl groups. Exemplary heterocyclic groups include pyrrolidinyl, tetrahydrofuranyl, tetrahydropyranyl, morpholinyl, thiomorpholinyl, piperidinyl, piperazinyl, thiazolidinyl, isothiazolidinyl, and imidazolidinyl.

The term "heterocyclylene" includes divalent radicals derived from the heterocyclyl groups described above.

The invention is inclusive of the compounds described herein in any of their pharmaceutically acceptable forms, including isomers such as diastereomers and enantiomers, salts, solvates, polymorphs, and the like. In particular, if a compound

is optically active, the invention specifically includes each of the compound's enantiomers as well as racemic mixtures of the enantiomers.

Preparation of the Compounds

Compounds of the invention can be prepared according to Reaction Scheme I where R_1', R_2, R, X, Z, and n are as defined above. In step (1) of Reaction Scheme I, a 1*H*-imidazo[4,5-*c*]quinolin-1-yl alcohol of Formula X is treated with *N*-hydroxyphthalimide under Mitsunobu reaction conditions to provide an *N*-phthalimide-protected 1*H*-imidazo[4,5-*c*]quinolin-1-yl hydroxylamine of Formula XI. The reaction is conveniently carried out by adding triphenylphosphine and *N*-hydroxyphthalimide to a solution of the alcohol of Formula X in a suitable solvent such as tetrahydrofuran and then slowly adding diisopropyl azodicarboxylate. The reaction can be carried out at ambient temperature or at an elevated temperature, such as 60°C. The product can be isolated using conventional methods. Many compounds of Formula X are known; see for example, U.S. Patent 4,689,338 (Gerster). Others can be readily prepared using known synthetic routes; see for example, U.S. Patent 5,605,899 (Gerster et al.) and U.S. Patent 5,175,296 (Gerster).

In step (2) of Reaction Scheme I, an *N*-phthalimide-protected 1*H*-imidazo[4,5-*c*]quinolin-1-yl hydroxylamine of Formula XI is oxidized to provide a 1*H*-imidazo[4,5-*c*]quinoline-5*N*-oxide of Formula XII using a conventional oxidizing agent capable of forming *N*-oxides. The reaction is conveniently carried out by adding 3-chloroperoxybenzoic acid to a solution of a compound of Formula XI in a solvent such as chloroform or dichloromethane. The reaction can be carried out at ambient temperature. The product can be isolated using conventional methods.

In step (3) of Reaction Scheme I, a 1*H*-imidazo[4,5-*c*]quinoline-5*N*-oxide of Formula XII is aminated to provide a 1*H*-imidazo[4,5-*c*]quinolin-4-amine of Formula XIII. Step (3) involves the activation of an *N*-oxide of Formula XII by conversion to an ester and then reacting the ester with an aminating agent. Suitable activating agents include alkyl- or arylsulfonyl chlorides such as benzenesulfonyl

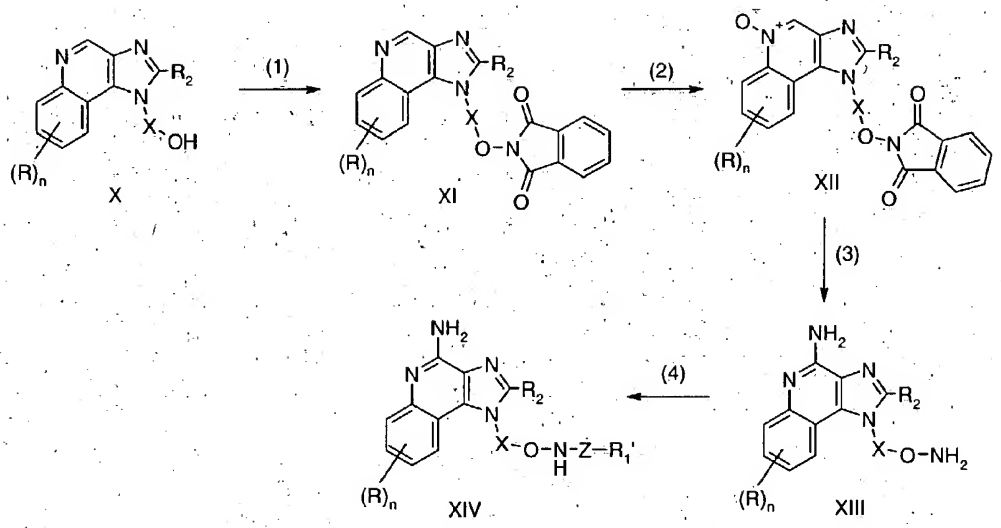
chloride, methanesulfonyl chloride, or *p*-toluenesulfonyl chloride. Suitable aminating agents include ammonia, in the form of ammonium hydroxide, for example, and ammonium salts such as ammonium carbonate, ammonium bicarbonate, and ammonium phosphate. The reaction is conveniently carried out by adding ammonium hydroxide to a solution of the *N*-oxide of Formula XII in a suitable solvent such as dichloromethane or chloroform and then adding *p*-toluenesulfonyl chloride. The reaction can be carried out at ambient temperature. Under these reaction conditions, the *N*-phthalimide protecting group is removed to provide the 1*H*-imidazo[4,5-*c*]quinolin-4-amine of Formula XIII, which represents a subgenus of the invention. The product or pharmaceutically acceptable salt thereof can be isolated from the reaction mixture using conventional methods.

In step (4) of Reaction Scheme I, a 1*H*-imidazo[4,5-*c*]quinolin-4-amine of Formula XIII is converted to a 1*H*-imidazo[4,5-*c*]quinolin-1-yl hydroxylamine of Formula XIV using conventional methods. For example, a 1*H*-imidazo[4,5-*c*]quinolin-4-amine of Formula XIII can react with an acid chloride of Formula $R_1'C(O)Cl$ or a sulfonyl chloride of Formula $R_1'S(O)_2Cl$ to provide a compound of Formula XIV in which Z is -C(O)- or $-S(O)_2-$, respectively. Numerous acid chlorides of Formula $R_1'C(O)Cl$ and sulfonyl chlorides of Formula $R_1'S(O)_2Cl$ are commercially available; others can be readily prepared using known synthetic methods. The reaction can be conveniently carried out by adding the acid chloride of Formula $R_1'C(O)Cl$ or sulfonyl chloride of Formula $R_1'S(O)_2Cl$ to a solution of the 1*H*-imidazo[4,5-*c*]quinolin-4-amine of Formula XIII and a base such as triethylamine or *N*,*N*-diisopropylethylamine in a suitable solvent such as chloroform or dichloroethane. The reaction can be carried out at ambient temperature or at an elevated temperature, such as 50°C. The product or pharmaceutically acceptable salt thereof can be isolated using conventional methods.

Ureas of Formula XIV, where Z is $-C(O)-N(R_5)-$, can be prepared by reacting a 1*H*-imidazo[4,5-*c*]quinolin-4-amine of Formula XIII with isocyanates of Formula $R_1'N=C=O$. Numerous isocyanates of Formula $R_1'N=C=O$ are commercially available; others can be readily prepared using known synthetic

methods. The reaction can be conveniently carried out by adding the isocyanate of Formula R_1'N=C=O to a solution of the 1*H*-imidazo[4,5-*c*]quinolin-4-amine of Formula XIII and a base such as triethylamine in a suitable solvent such as chloroform. The reaction can be carried out at ambient temperature or at an elevated temperature, such as 50°C. Alternatively, a compound of Formula XIII can be treated with a an isocyanate of Formula R_1'(CO)N=C=O, a thioisocyanate of Formula R_1'N=C=S, a sulfonyl isocyanate of Formula R_1'$S(O)_2$N=C=O, or a carbamoyl chloride of Formula R_1'$N(R_5)$-C(O)Cl. The product or pharmaceutically acceptable salt thereof can be isolated using conventional methods.

Reaction Scheme I



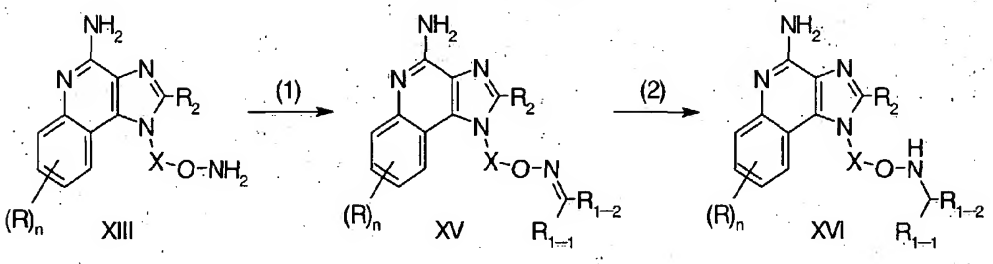
Compounds of the invention can be prepared according to Reaction Scheme II where R_2, R, X, and n are as defined above and $R_{1\text{-}1}$ and $R_{1\text{-}2}$ are independently selected from the group consisting of hydrogen, alkyl, alkenyl, aryl, alkylene-aryl, heteroaryl, heterocyclyl, and alkyl, alkenyl, aryl, arylalkylenyl, heteroaryl or heterocyclyl substituted by one or more substituents selected from the group consisting of OH, alkyl, haloalkyl, hydroxyalkyl, -O-alkyl, -S-alkyl, -O-haloalkyl,

halogen, nitrile, aryl, heteroaryl, heterocyclyl, -O-aryl, -O-alkylene-aryl, -C(O)-O-alkyl,-C(O)-N(R_5)$_2$ and -N(R_5)-C(O)-alkyl; or R_{1-1} and R_{1-2} can join together to form a ring system containing one or two saturated or unsaturated rings optionally including one or more heteroatoms.

In step (1) of Reaction Scheme II, a 1*H*-imidazo[4,5-*c*]quinolin-4-amine of Formula XIII reacts with an aldehyde or ketone of Formula $R_{1-1}C(O)R_{1-2}$ to provide a 1*H*-imidazo[4,5-*c*]quinolin-1-yl oxime of Formula XV. Numerous aldehydes and ketones of Formula $R_{1-1}C(O)R_{1-2}$ are commercially available; others can be readily prepared using known synthetic methods. The reaction can be conveniently carried out by adding the aldehyde or ketone of Formula $R_{1-1}C(O)R_{1-2}$ to a solution of the 1*H*-imidazo[4,5-*c*]quinolin-4-amine of Formula XIII in a suitable solvent such as methanol. The reaction can be carried out at ambient temperature. The product or pharmaceutically acceptable salt thereof can be isolated using conventional methods.

In step (2) of Reaction Scheme II, a 1*H*-imidazo[4,5-*c*]quinolin-1-yl oxime of Formula XV is reduced to provide a 1*H*-imidazo[4,5-*c*]quinolin-1-yl hydroxylamine of Formula XVI, which represents a subgenus of the invention. The reduction is conveniently carried out by treating the 1*H*-imidazo[4,5-*c*]quinolin-1-yl oxime of Formula XV with excess sodium cyanoborohydride in a suitable solvent or solvent mixture such as methanol/acetic acid. The reaction can be carried out at ambient temperature. The product or pharmaceutically acceptable salt thereof can be isolated using conventional methods.

Reaction Scheme II



Pharmaceutical Compositions and Biological Activity

Pharmaceutical compositions of the invention contain an effective amount of a compound of the invention as described above in combination with a pharmaceutically acceptable carrier.

The terms "a therapeutically effective amount" and "an effective amount" mean an amount of the compound sufficient to induce a therapeutic or prophylactic effect, such as cytokine induction, immunomodulation, antitumor activity, and/or antiviral activity. Although the exact amount of active compound used in a pharmaceutical composition of the invention will vary according to factors known to those of skill in the art, such as the physical and chemical nature of the compound, the nature of the carrier, and the intended dosing regimen, it is anticipated that the compositions of the invention will contain sufficient active ingredient to provide a dose of about 100 nanograms per kilogram (ng/kg) to about 50 milligrams per kilogram (mg/kg), preferably about 10 micrograms per kilogram (μg/kg) to about 5 mg/kg, of the compound to the subject. A variety of dosage forms may be used, such as tablets, lozenges, capsules, parenteral formulations, syrups, creams, ointments, aerosol formulations, transdermal patches, transmucosal patches and the like.

The compounds of the invention can be administered as the single therapeutic agent in the treatment regimen, or the compounds of the invention may be administered in combination with one another or with other active agents, including additional immune response modifiers, antivirals, antibiotics, antibodies, proteins, peptides, oligonucleotides, etc.

The compounds of the invention have been shown to induce the production of certain cytokines in experiments performed according to the tests set forth below. These results indicate that the compounds are useful as immune response modifiers that can modulate the immune response in a number of different ways, rendering them useful in the treatment of a variety of disorders.

Cytokines whose production may be induced by the administration of compounds according to the invention generally include interferon-α (IFN-α) and/or tumor necrosis factor-α (TNF-α) as well as certain interleukins (IL). Cytokines whose biosynthesis may be induced by compounds of the invention include IFN-α, TNF-α, IL-1, IL-6, IL-10 and IL-12, and a variety of other cytokines. Among other effects, these and other cytokines can inhibit virus production and tumor cell growth, making the compounds useful in the treatment of viral diseases and neoplastic diseases. Accordingly, the invention provides a method of inducing cytokine biosynthesis in an animal comprising administering an effective amount of a compound or composition of the invention to the animal. The animal to which the compound or composition is administered for induction of cytokine biosynthesis may have a disease as described *infra*, for example a viral disease or a neoplastic disease, or the animal may not have a disease, but instead be given the compound or composition for prophylaxis of a disease, for example a viral disease or a neoplastic disease. For prophylaxis of a disease, the compound or composition may be administered, for example, as a vaccine adjuvant. Thus, treatment may be therapeutic and/or prophylactic.

In addition to the ability to induce the production of cytokines, the compounds of the invention affect other aspects of the innate immune response. For example, natural killer cell activity may be stimulated, an effect that may be due to cytokine induction. The compounds may also activate macrophages, which in turn stimulate secretion of nitric oxide and the production of additional cytokines. Further, the compounds may cause proliferation and differentiation of B-lymphocytes.

Compounds of the invention also have an effect on the acquired immune response. For example, the production of the T helper type 1 (Th1) cytokine IFN-γ is induced indirectly and the production of the T helper type 2 (Th2) cytokines IL-4, IL-5 and IL-13 are inhibited upon administration of the compounds.

Diseases for which immune response modifiers (IRMs) identified herein may be used as treatments include, but are not limited to:

(a) viral diseases, such as genital warts, common warts, plantar warts, hepatitis B, hepatitis C, herpes simplex virus type I and type II, molluscum contagiosum, variola, HIV, CMV, VZV, rhinovirus, adenovirus, coronavirus, influenza, and para-influenza;

(b) bacterial diseases, such as tuberculosis, mycobacterium avium, and leprosy;

(c) other infectious diseases, such as fungal diseases, chlamydia, candida, aspergillus, cryptococcal meningitis, pneumocystis carnii, cryptosporidiosis, histoplasmosis, toxoplasmosis, trypanosome infection, and leishmaniasis;

(d) neoplastic diseases, such as intraepithelial neoplasias, cervical dysplasia, actinic keratosis, basal cell carcinoma, squamous cell carcinoma, hairy cell leukemia, Karposi's sarcoma, melanoma, renal cell carcinoma, myelogeous leukemia, multiple myeloma, non-Hodgkin's lymphoma, cutaneous T-cell lymphoma, and other cancers;

(e) Th2 mediated, atopic, and autoimmune diseases, such as atopic dermatitis or eczema, eosinophilia, asthma, allergy, allergic rhinitis, systemic lupus erythematosis, essential thrombocythaemia, multiple sclerosis, Ommen's syndrome, discoid lupus, alopecia areata, inhibition of keloid formation and other types of scarring, and enhancing would healing, including chronic wounds; and

(f) as a vaccine adjuvant for use in conjunction with any material that raises either humoral and/or cell mediated immune response, such live viral and bacterial immunogens and inactivated viral, tumor-derived, protozoal, organism-derived, fungal, and bacterial immunogens, toxoids, toxins, polysaccharides, proteins, glycoproteins, peptides, cellular vaccines, DNA vaccines, recombinant proteins, glycoproteins, and peptides, and the like, for use in connection with, e.g., BCG, cholera, plague, typhoid, hepatitis A, B, and C, influenza A and B, parainfluenza, polio, rabies, measles, mumps, rubella, yellow fever, tetanus, diphtheria, hemophilus influenza b, tuberculosis, meningococcal and pneumococcal vaccines,

adenovirus, HIV, chicken pox, cytomegalovirus, dengue, feline leukemia, fowl plague, HSV-1 and HSV-2, hog cholera, Japanese encephalitis, respiratory syncytial virus, rotavirus, papilloma virus, and yellow fever.

IRMs may also be particularly helpful in individuals having compromised immune function. For example, IRM compounds may be used for treating the opportunistic infections and tumors that occur after suppression of cell mediated immunity in, for example, transplant patients, cancer patients and HIV patients.

Thus, one or more of the above diseases or types of diseases, for example, a viral disease or a neoplastic disease may be treated in an animal in need there of (having the disease) by administering an effective amount of a compound or salt of formula (I) to the animal.

An amount of a compound effective to induce cytokine biosynthesis is an amount sufficient to cause one or more cell types, such as monocytes, macrophages, dendritic cells and B-cells to produce an amount of one or more cytokines such as, for example, IFN-α, TNF-α, IL-1, IL-6, IL-10 and IL-12 that is increased over the background level of such cytokines. The precise amount will vary according to factors known in the art but is expected to be a dose of about 100 ng/kg to about 50 mg/kg, preferably about 10 μg/kg to about 5 mg/kg. The invention also provides a method of treating a viral infection in an animal and a method of treating a neoplastic disease in an animal comprising administering an effective amount of a compound or composition of the invention to the animal. An amount effective to treat or inhibit a viral infection is an amount that will cause a reduction in one or more of the manifestations of viral infection, such as viral lesions, viral load, rate of virus production, and mortality as compared to untreated control animals. The precise amount that is effective for such treatment will vary according to factors known in the art but is expected to be a dose of about 100 ng/kg to about 50 mg/kg, preferably about 10 μg/kg to about 5 mg/kg. An amount of a compound effective to treat a neoplastic condition is an amount that will cause a reduction in tumor size or in the number of tumor foci. Again, the precise amount will vary according to

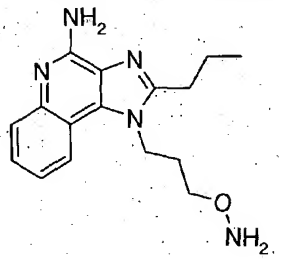
factors known in the art but is expected to be a dose of about 100 ng/kg to about 50 mg/kg, preferably about 10 μg/kg to about 5 mg/kg.

Objects and advantages of this invention are further illustrated by the following examples, but the particular materials and amounts thereof recited in these examples, as well as other conditions and details, should not be construed to unduly limit this invention.

EXAMPLES

Example 1

O-[3-(4-Amino-2-propyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)propyl]hydroxylamine



Part A

A solution of 1-(3-hydroxypropyl)-2-propyl-1*H*-imidazo[4,5-*c*]quinoline (20.0 grams (g), 74.3 millimoles (mmol)) in tetrahydrofuran (300 milliliters (mL)) was cooled to approximately 0°C; triphenylphosphine (23.4 g, 89.1 mmol) and *N*-hydroxyphthalimide (14.5 g, 89.1 mmol) were then added. After five minutes of stirring, diisopropyl azodicarboxylate (17.5 mL, 89.1 mmol) was added dropwise over a period of 15 minutes. The reaction was allowed to warm to room temperature and stirred overnight. The solvent was removed under reduced pressure, and the residue was dissolved in chloroform (300 mL). A solution of hydrochloric acid (150 mL of 6 molar (M)) was then added, and approximately 50 mL of the solvent was removed under reduced pressure to provide a white precipitate, which was stirred for ten minutes and isolated by filtration. Additional

salt eventually precipitated from the filtrate and was isolated by filtration. Chloroform (300 mL) and water (300 mL) were added to the salt, and solid sodium bicarbonate was added to the mixture to adjust to pH 8. The organic solution was then dried over magnesium sulfate, filtered, and concentrated under reduced pressure to provide 28.4 g of 2-[3-(2-propyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)propoxy]isoindole-1,3-dione as a white solid.

^{1}H NMR (300 MHz, $CDCl_3$) δ 9.3 (s, 1H), 8.3 (m, 2H), 7.9 (m, 2H), 7.8 (m, 2H), 7.6 (m, 2H), 5.0 (t, J = 7.3 Hz, 2H), 4.4 (t, J = 5.3 Hz, 2H), 3.1 (t, J = 7.5 Hz, 2H), 2.4 (m, 2H), 2.1 (br s, m, 4H), 1.2 (t, J = 7.3 Hz, 3H);

MS (APCI) *m/z* 415 $(M + H)^+$.

Part B

3-Chloroperoxybenzoic acid (14.9 g, 66.4 mmol) (mCPBA, available as an approximately 77% pure mixture) was added to a solution of 2-[3-(2-propyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)propoxy]isoindole-1,3-dione (25.0 g, 60.3 mmol) in chloroform (200 mL), and the reaction was stirred for seven hours at room temperature. An analysis by liquid chromatography/mass spectrometry (LC/MS) indicated that the reaction was incomplete, and additional mCPBA (4.96 g, 22.1 mmol) was added. The reaction was allowed to stir at room temperature overnight. The solution was then washed with brine (2 x 100 mL) and saturated aqueous sodium bicarbonate (2 x 100 mL), dried over magnesium sulfate, filtered, and concentrated under reduced pressure to provide a fluffy, light-brown solid. The solid was dried under high vacuum for one hour to provide 25.7 g of 2-[3-(5-oxido-2-propyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)propoxy]isoindole-1,3-dione as a white solid.

^{1}H NMR (300 MHz, $CDCl_3$) δ 9.1 (m, 2H), 8.3 (m, 1H), 7.9-7.7 (m, 6H), 5.0 (t, J = 7.4 Hz, 2H), 4.4 (t, J = 5.3 Hz, 2H), 3.1 (t, J = 7.5 Hz, 2H), 2.4 (m, 2H), 2.1 (br s, m, 4H), 1.2 (t, J = 7.3 Hz, 3H);

MS (APCI) *m/z* 431 $(M + H)^+$.

Part C

Ammonium hydroxide (75 mL) and *p*-toluenesulfonyl chloride (4.87 g, 25.6 mmol) were added to a solution of 2-[3-(5-oxido-2-propyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)propoxy]isoindole-1,3-dione (10.0 g, 23.2 mmol) in chloroform (100 mL), and the resulting mixture was stirred vigorously for one hour. A white precipitate was removed by filtration, and the filtrate layers were separated. The organic solution was washed with brine (2 x 150 mL), dried over magnesium sulfate, filtered, and concentrated under reduced pressure to provide a yellow solid. The solid was purified by column chromatography on silica gel (eluting with dichloromethane:methanol:ammonium hydroxide ranging in ratios from 94:5:1 to 91:8:1) to provide 4.31 g of *O*-[3-(4-amino-2-propyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)propyl]hydroxylamine as a beige powder, melting point (mp) 145-148°C.

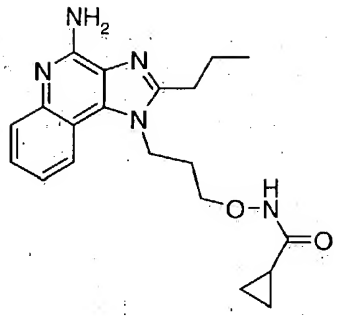
^{1}H NMR (300 MHz, DMSO-d_6) δ 8.1 (d, J = 7.5 Hz, 1H), 7.6 (d, J = 8.3 Hz, 1H), 7.4 (t, J = 8.1 Hz, 1H), 7.3 (t, J = 8.1 Hz, 1H), 6.5 (br s, 2H), 6.1 (br s, 2H), 4.6 (t, J = 7.2 Hz, 2H), 3.6 (t, J = 5.6 Hz, 2H), 2.9 (t, J = 7.4 Hz, 2H), 2.1 (m, 2H), 1.9 (m, 2H), 1.1 (t, J = 7.3 Hz, 3H); ^{13}C NMR (75 MHz, DMSO-d_6) δ 153.4, 152.0, 145.0, 132.6, 126.8, 126.6, 121.5, 120.4, 115.1, 71.6, 42.5, 29.2, 28.5, 21.3, 14.2;

MS (APCI) *m/z* 300 $(M + H)^+$;

Anal. calcd for $C_{16}H_{21}N_5O$: C, 64.19; H, 7.07; N, 23.39. Found: C, 63.94; H, 7.20; N, 23.11.

Example 2

Cyclopropanecarboxylic acid [3-(4-amino-2-propyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)propoxy]amide



Triethylamine (1.86 mL, 13.4 mmol) was added to a mixture of *O*-[3-(4-amino-2-propyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)propyl]hydroxylamine (2.00 g, 6.68 mmol), prepared as described in Example 1, and chloroform (20 mL). Cyclopropanecarbonyl chloride (673 µL, 7.35 mmol) was then added, and the resulting solution was stirred at room temperature overnight. The reaction was diluted with chloroform (50 mL), washed with brine (2 x 100 mL) and saturated aqueous sodium bicarbonate (2 x 100 mL), dried over magnesium sulfate, filtered, and concentrated under reduced pressure. The crude product was purified twice by column chromatography on silica gel (eluting sequentially with 97:2:1 dichloromethane:methanol:ammonium hydroxide and 96:3:1 dichloromethane:methanol:ammounium hydroxide) to provide 532 mg of cyclopropanecarboxylic acid [3-(4-amino-2-propyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)propoxy]amide as a white powder, mp 103-105°C.

^{1}H NMR (300 MHz, DMSO-d_6) δ 8.0 (d, *J* = 7.9 Hz, 1H), 7.8 (d, *J* = 7.8 Hz, 1H), 7.5 (t, *J* = 7.4 Hz, 1H), 7.3 (t, *J* = 8.0 Hz, 1H), 5.4 (br s, 2H), 4.7 (m, 2H), 4.0 (t, *J* = 5.4 Hz, 2H), 2.9 (t, *J* = 7.2 Hz, 2H), 2.2 (m, 2H), 1.9 (m, 2H), 1.3 (m, 1H), 1.1 (m, 5H), 0.9 (m, 2H);

MS (APCI) *m/z* 368 $(M + H)^+$;

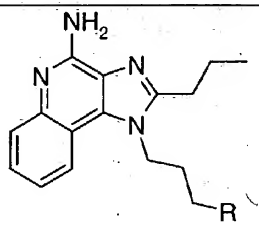
HRMS (ESI) Theoretical mass: 368.2087, measured mass: 368.2073.

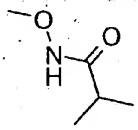
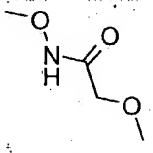
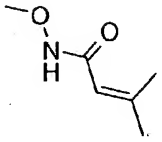
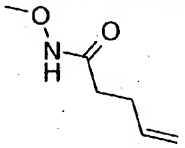
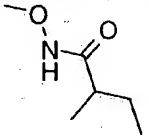
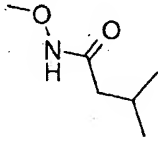
Anal. calcd for $C_{20}H_{25}N_5O_2 \cdot 0.3CH_2Cl_2$: C, 62.05; H, 6.57; N, 17.82. Found: C, 61.85; H, 6.68; N, 17.79.

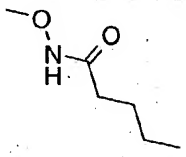
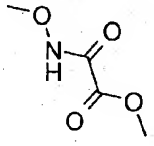
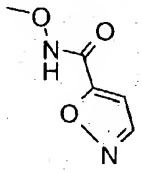
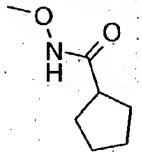
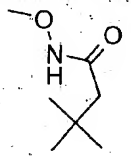
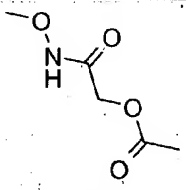
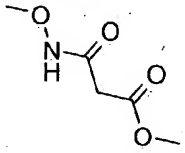
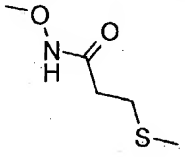
Examples 3-70

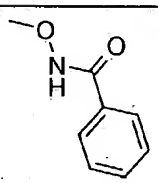
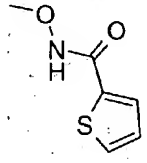
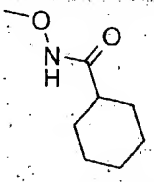
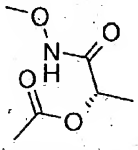
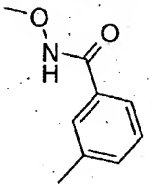
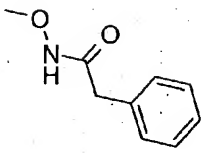
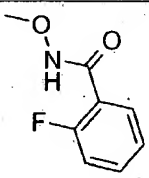
An acid chloride from the table below (0.83 equivalents, 0.057 mmol) was added to a test tube containing a solution of *O*-[3-(4-amino-2-propyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)propyl]hydroxylamine (21 mg, 0.070 mmol) and *N*,*N*-diisopropylethylamine (24 microliters (μL), 0.14 mmol) in chloroform (2 mL). In Examples 17 and 69, dichloroethane was used as the solvent instead of chloroform. The test tube was capped and placed on a shaker at ambient temperature overnight (approximately 18 hours). For Example 70, an analysis by LC/MS indicated that the reaction was incomplete; therefore for Examples 69 and 70, the solutions additionally were heated at 50°C for seven hours. The solvent was removed from the test tubes by vacuum centrifugation. The compounds were purified by preparative high performance liquid chromatography (prep HPLC) using a Waters Fraction Lynx automated purification system. The prep HPLC fractions were analyzed using a Micromass LC-TOFMS, and the appropriate fractions were centrifuge evaporated to provide the trifluoroacetate salt of the desired compound. Column: Phenomenex Luna C18(2), 21.2 x 50 millimeters (mm), 10 micron particle size, 100 Angstroms (Å) pore; flow rate: 25 mL/min; non-linear gradient elution from 5-95% B in 9 minutes (min), then hold at 95% B for 2 min, where A is 0.05% trifluoroacetic acid/water and B is 0.05% trifluoroacetic acid/acetonitrile; fraction collection by mass-selective triggering. The table below shows the acid chloride used for each example, the structure of the resulting compound, and the observed accurate mass for the isolated trifluoroacetate salt.

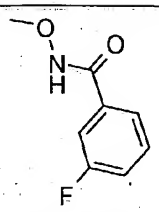
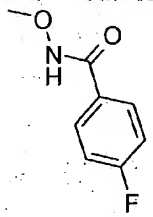
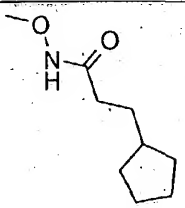
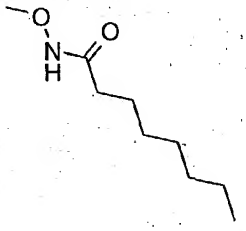
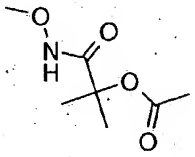
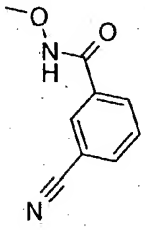
Examples 3-70

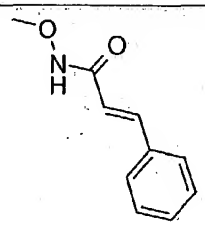
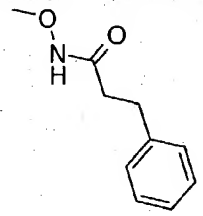
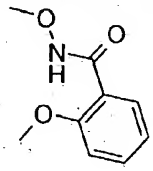
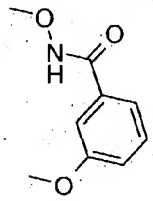
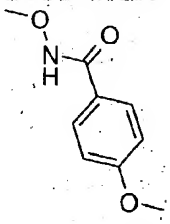
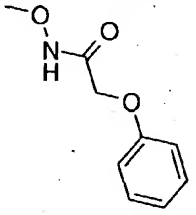


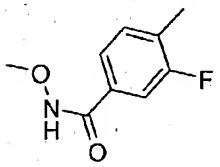
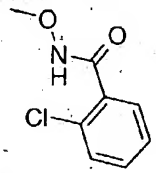
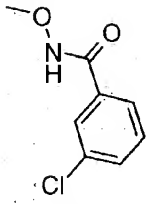
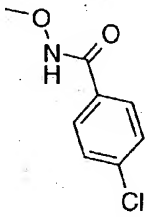
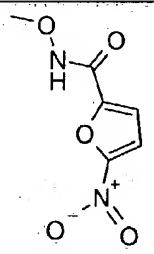
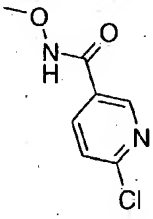
Example	Acid Chloride	R	Measured Mass (M+H)
3	Isobutryl chloride		370.2252
4	Methoxyacetyl chloride		372.2035
5	3,3-Dimethylacryloyl chloride		382.2262
6	4-Pentenoyl chloride		382.2228
7	2-Methylbutryl chloride		384.2391
8	Isovaleryl chloride		384.2418

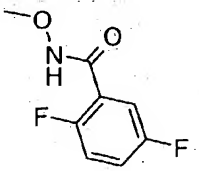
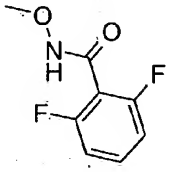
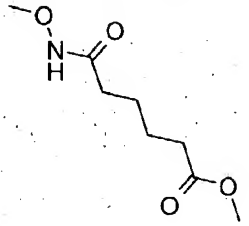
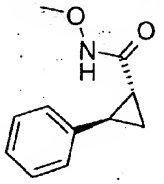
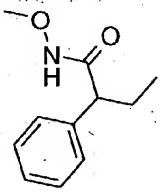
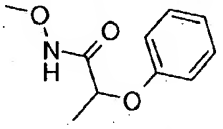
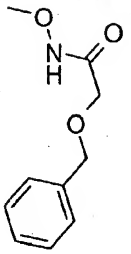
9	Pentanoyl chloride		384.2419
10	Methyloxalyl chloride		386.1835
11	Isoxazole-5-carbonyl chloride		395.1826
12	Cyclopentanecarbonyl chloride		396.2420
13	*tert*-Butylacetyl chloride		398.2581
14	Acetoxyacetyl chloride		400.1979
15	Methylmalonyl chloride		400.1965
16	3-Methylthiopropionyl chloride		402.1950

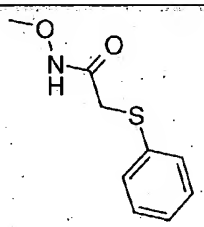
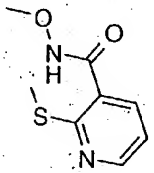
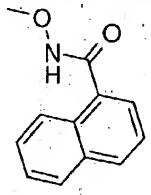
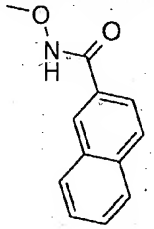
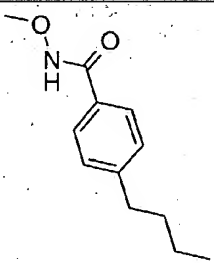
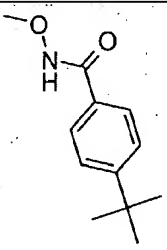
17	Benzoyl chloride		404.2097
18	Thiophene-2-carbonyl chloride		410.1679
19	Cyclohexanecarbonyl chloride		410.2553
20	(*S*)-(-)-2-Acetoxypropionyl chloride		414.2162
21	*m*-Toluoyl chloride		418.2261
22	Phenylacetyl chloride		418.2253
23	2-Fluorobenzoyl chloride		422.2006

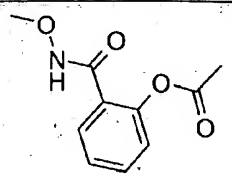
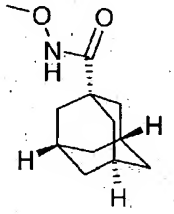
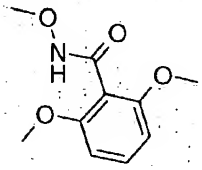
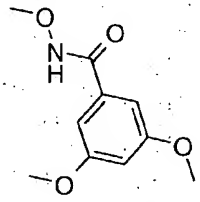
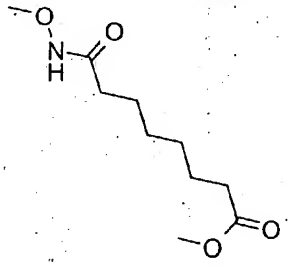
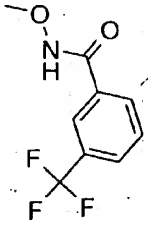
24	3-Fluorobenzoyl chloride		422.2020
25	4-Fluorobenzoyl chloride		422.1982
26	3-Cyclopentylpropionyl chloride		424.2731
27	Octanoyl chloride		426.2857
28	2-Acetoxyisobutyryl chloride		428.2306
29	3-Cyanobenzoyl chloride		429.2063

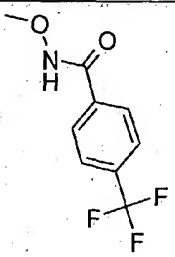
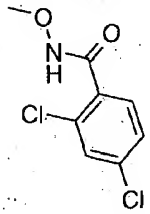
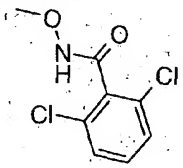
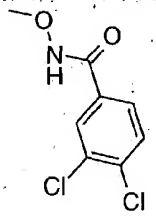
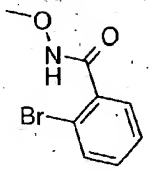
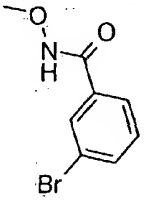
30	Cinnamoyl chloride		430.2243
31	Hydrocinnamoyl chloride		432.2374
32	2-Methoxybenzoyl chloride		434.2227
33	*m*-Anisoyl chloride		434.2213
34	*p*-Anisoyl chloride		434.2217
35	Phenoxyacetyl chloride		434.2197

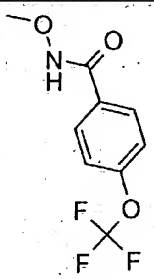
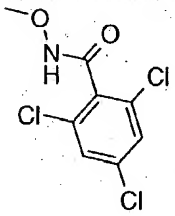
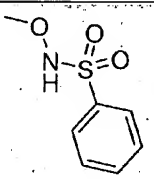
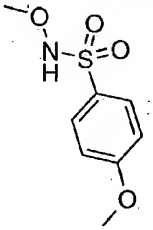
36	3-Fluoro-4-methylbenzoyl chloride		436.2164
37	2-Chlorobenzoyl chloride		438.1688
38	3-Chlorobenzoyl chloride		438.1714
39	4-Chlorobenzoyl chloride		438.1705
40	5-Nitro-2-furoyl chloride		439.1762
41	6-Chloronicotinyl chloride		439.1685

42	2,5-Difluorobenzoyl chloride		440.1900
43	2,6-Difluorobenzoyl chloride		440.1910
44	Methyladipoyl chloride		442.2446
45	*trans*-2-Phenyl-1-cyclopropanecarbonyl chloride		444.2380
46	2-Phenylbutyryl chloride		446.2541
47	2-Phenoxypropionyl chloride		448.2361
48	Benzyloxyacetyl chloride		448.2356

49	(Phenylthio)acetyl chloride		450.1978
50	2-(Methylthio)nicotinyl chloride		451.1892
51	1-Naphthoyl chloride		454.2273
52	2-Naphthoyl chloride		454.2226
53	4-*n*-Butylbenzoyl chloride		460.2684
54	4-*tert*-Butylbenzoyl chloride		460.2740

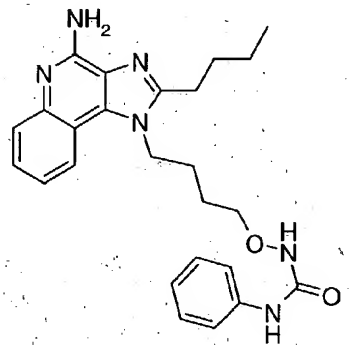
55	*O*-Acetylsalicyloyl chloride		462.2137
56	1-Adamantanecarbonyl chloride		462.2875
57	2,6-Dimethoxybenzoyl chloride		464.2310
58	3,5-Dimethoxybenzoyl chloride		464.2317
59	Methyl 8-chloro-8-oxooctanoate		470.2797
60	3-(Trifluoromethyl)-benzoyl chloride		472.1980

61	4-(Trifluoromethyl)-benzoyl chloride		472.1990
62	2,4-Dichlorobenzoyl chloride		472.1285
63	2,6-Dichlorobenzoyl chloride		472.1296
64	3,4-Dichlorobenzoyl chloride		472.1337
65	2-Bromobenzoyl chloride		482.1191
66	3-Bromobenzoyl chloride		482.1184

67	4-(Trifluoromethoxy)-benzoyl chloride		488.1936
68	2,4,6-Trichlorobenzoyl chloride		506.0902
69	Benzenesulfonyl chloride		440.1761
70	4-Methoxybenzenesulfonyl chloride		470.1863

Example 71

N-{*O*-[4-(4-Amino-2-butyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)butyl]hydroxyl}-*N'*-phenylurea



Part A

Triphenylphosphine (21.2 g, 80.7 mmol) and *N*-hydroxyphthalimide (13.2 g, 80.7 mmol) were added to a solution of 2-butyl-1-(4-hydroxybutyl)-1*H*-imidazo[4,5-*c*]quinoline (16.0 g, 53.8 mmol) in tetrahydrofuran (200 mL). The mixture was stirred for five minutes and then was cooled to approximately 0°C. Diisopropyl azodicarboxylate (19.6 g, 96.8 mmol) was added dropwise, and the reaction was allowed to warm to room temperature and stirred for three hours. An analysis by LC/MS indicated the presence of starting material, and the reaction was stirred at 60°C overnight. An analysis by LC/MS indicated the presence of starting material, and additional triphenylphosphine, *N*-hydroxyphthalimide, and diisopropyl azodicarboxylate (26.9 mmol of each) were added to the reaction mixture. The reaction was stirred at room temperature for two hours and heated at reflux for three hours. The reaction was concentrated under reduced pressure, and the residue was dissolved in chloroform (200 mL). The resulting solution was washed with brine (3 x 150 mL), dried over magnesium sulfate, filtered through a layer of CELITE filter aid, and concentrated under reduced pressure. An analysis of the crude product mixture by LC/MS indicated that starting material was still present. The mixture was dissolved in tetrahydrofuran (200 mL) and treated with triphenylphosphine (21.2 g, 80.7 mmol), *N*-hydroxyphthalimide (13.2 g, 80.7 mmol), and diisopropyl

azodicarboxylate (19.6 g, 96.8 mmol) as described above. The reaction was stirred overnight at room temperature. The product was present as a white precipitate, which was isolated by filtration and washed with tetrahydrofuran to provide 8.68 g of 2-[4-(2-butyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)butoxy]isoindole-1,3-dione as a white solid.

^{1}H NMR (300 MHz, $CDCl_3$) δ 9.3 (s, 1H), 8.3 (m, 2H), 7.9 (m, 2H), 7.8 (m, 2H), 7.7 (m, 2H), 4.7 (t, J = 7.9 Hz, 2H), 4.3 (t, J = 5.8 Hz, 2H), 3.1 (t, J = 7.6 Hz, 2H), 2.3 (m, 2H), 2.0 (m, 4H), 1.6 (m, 2H), 1.1 (t, J = 7.3 Hz, 3H);

MS (APCI) *m/z* 443 $(M + H)^+$.

Part B

A solution of 2-[4-(2-butyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)butoxy]isoindole-1,3-dione (7.65 g, 17.3 mmol) in dichloromethane (100 mL) was treated with mCPBA (4.65 g, 20.7 mmol), and the resulting orange solution was stirred for four hours at room temperature. The solution was then diluted with dichloromethane (100 mL), washed with brine (3 x 100 mL), dried over magnesium sulfate, filtered through a layer of CELITE filter aid, and concentrated under reduced pressure to provide 9.92 g of 2-[4-(2-butyl-5-oxido-1*H*-imidazo[4,5-*c*]quinolin-1-yl)butoxy]isoindole-1,3-dione as a red semi-solid.

Part C

A mixture of 2-[4-(2-butyl-5-oxido-1*H*-imidazo[4,5-*c*]quinolin-1-yl)butoxy]isoindole-1,3-dione (8.92 g, 19.5 mmol) in dichloroethane (100 mL) was shaken vigorously until it became homogeneous. With vigorous stirring, ammonium hydroxide (100 mL) and *p*-toluenesulfonyl chloride (4.45 g, 23.4 mmol) were added sequentially. The reaction was stirred overnight at room temperature. The product was present as a white precipitate, which was isolated by filtration to provide 1.97 g of *O*-[4-(4-amino-2-butyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)butyl]hydroxylamine as a white solid.

^{1}H NMR (300 MHz, $CDCl_3$) δ 8.0 (d, *J* = 8.2 Hz, 1H), 7.8 (d, *J* = 8.3 Hz, 1H), 7.5 (t, *J* = 7.1 Hz, 1H), 7.3 (t, *J* = 7.1 Hz, 1H), 5.6 (br s, 2H), 5.2 (br s, 2H), 4.5 (t, *J* = 7.8 Hz, 2H), 3.8 (t, *J* = 6.2 Hz, 2H), 2.9 (t, *J* = 7.6 Hz, 2H), 1.7-2.0 (m, 6H), 1.6 (m, 2H), 1.0 (t, *J* = 7.3 Hz, 3H);

MS (APCI) *m/z* 328 $(M + H)^+$.

The filtrate with diluted with chloroform, washed with brine (3 x 100 mL), dried over magnesium sulfate, filtered through a layer of CELITE filter aid, and concentrated under reduced pressure to provide 5.72 g additional product as a red semi-solid.

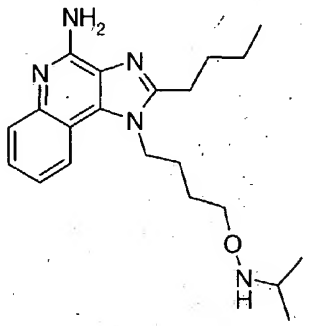
Part D

Triethylamine (495 µL, 3.55 mmol) and phenyl isocyanate (316 mg, 1.65 mmol) were added to a mixture of *O*-[4-(4-amino-2-butyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)butyl]hydroxylamine (580 mg, 1.77 mmol) in chloroform (10 mL). The reaction was heated at 50°C overnight and became homogeneous. The solution was diluted with chloroform, washed with brine (2x), dried over magnesium sulfate, filtered through a layer of CELITE filter aid, and concentrated under reduced pressure to provide the crude product as an orange solid (700 mg). The crude product was purified by column chromatography on silica gel (eluting with dichloromethane:methanol in ratios ranging from 100:0 to 95:5) to provide 90 milligrams (mg) of *N*-{*O*-[4-(4-amino-2-butyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)butyl]hydroxyl}-*N'*-phenylurea as a white solid.

HRMS (ESI) Theoretical mass: 447.2508, measured mass: 447.2497.

Example 72

O-[4-(4-Amino-2-butyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)butyl]-*N*-(2-propyl)hydroxylamine



Part A

Acetone (444 mg, 7.65 mmol) was added to a solution of *O*-[4-(4-amino-2-butyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)butyl]hydroxylamine (0.500 g, 1.53 mmol), prepared as described in Parts A-C of Example 71, in methanol (7 mL), and the reaction was stirred overnight at room temperature. The solvent was removed under reduced pressure and then further dried under high vacuum to provide 358 mg of propan-2-one *O*-[4-(4-amino-2-butyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)butyl]oxime as a white solid, mp 115-117°C.

^{1}H NMR (300 MHz, DMSO-d_6) δ 8.0 (d, *J* = 7.8 Hz, 1H), 7.7 (d, *J* = 8.3 Hz, 1H), 7.5 (t, *J* = 8.0 Hz, 1H), 7.3 (t, *J* = 8.1 Hz, 1H), 6.5 (br s, 2H), 4.5 (t, *J* = 7.2 Hz, 2H), 4.0 (t, *J* = 6.0 Hz, 2H), 2.9 (t, *J* = 7.5 Hz, 2H), 1.9-1.6 (m, 12H), 1.5 (m, 2H), 1.1 (t, *J* = 7.3 Hz, 3H);

^{13}C NMR (75 MHz, DMSO-d_6) δ 154.2, 153.4, 152.0, 144.8, 132.6, 128.4, 126.6, 126.4, 121.5, 120.3, 115.1, 71.9, 44.9, 30.0, 26.8, 26.5, 25.9, 22.3, 21.6, 15.4, 14.1;

MS (APCI) *m/z* 368 $(M + H)^+$;

HRMS (ESI) Theoretical mass: 368.2469, measured mass: 368.2450.

Part B

Sodium cyanoborohydride (2 mL of a 1M solution in tetrahydrofuran) was added to a mixture of propan-2-one *O*-[4-(4-amino-2-butyl-1*H*-imidazo[4,5-

c]quinolin-1-yl)butyl]oxime (358 mg, 0.974 mmol), methanol (5 mL), and acetic acid (2 mL). The reaction was stirred overnight at ambient temperature. A precipitate was present and was removed by filtration. The filtrate was diluted with chloroform, and the resulting solution was washed twice with brine, dried over magnesium sulfate, filtered through a layer of CELITE filter aid, and concentrated under reduced pressure. The crude product was purified by column chromatography on silica gel to provide 100 mg of *O*-[4-(4-amino-2-butyl-1*H*-imidazo[4,5-*c*]quinolin-1-yl)butyl]-*N*-(2-propyl)hydroxylamine as an off-white solid. HRMS (ESI) Theoretical mass: 370.2607, measured mass: 370.2600.

CYTOKINE INDUCTION IN HUMAN CELLS

Compounds of the invention have been found to induce cytokine biosynthesis when tested using the method described below.

An in vitro human blood cell system is used to assess cytokine induction. Activity is based on the measurement of interferon and tumor necrosis factor (α) (IFN and TNF, respectively) secreted into culture media as described by Testerman et. al. In "Cytokine Induction by the Immunomodulators Imiquimod and S-27609," *Journal of Leukocyte Biology*, *58*, 365-372 (September, 1995).

Blood Cell Preparation for Culture

Whole blood from healthy human donors is collected by venipuncture into EDTA vacutainer tubes. Peripheral blood mononuclear cells (PBMC) are separated from whole blood by density gradient centrifugation using HISTOPAQUE-1077. Blood is diluted 1:1 with Dulbecco's Phosphate Buffered Saline (DPBS) or Hank's Balanced Salts Solution (HBSS). The PBMC layer is collected and washed twice with DPBS or HBSS and resuspended at 4 x 10^6 cells/mL in RPMI complete. The PBMC suspension is added to 48 well flat bottom sterile tissue culture plates (Costar, Cambridge, MA or Becton Dickinson Labware, Lincoln Park, NJ) containing an equal volume of RPMI complete media containing test compound.

Compound Preparation

The compounds are solubilized in dimethyl sulfoxide (DMSO). The DMSO concentration should not exceed a final concentration of 1% for addition to the culture wells. The compounds are generally tested at concentrations ranging from 30-0.014 micromolar (μM).

Incubation

The solution of test compound is added at 60 μM to the first well containing RPMI complete and serial 3 fold dilutions are made in the wells. The PBMC suspension is then added to the wells in an equal volume, bringing the test compound concentrations to the desired range (30-0.014 μM). The final concentration of PBMC suspension is 2 x 10^6 cells/mL. The plates are covered with sterile plastic lids, mixed gently and then incubated for 18 to 24 hours at 37°C in a 5% carbon dioxide atmosphere.

Separation

Following incubation the plates are centrifuged for 10 minutes at 1000 rpm (approximately 200 x g) at 4°C. The cell-free culture supernatant is removed with a sterile polypropylene pipet and transferred to sterile polypropylene tubes. Samples are maintained at -30°C to -70°C until analysis. The samples are analyzed for interferon (α) by ELISA and for tumor necrosis factor (α) by ELISA or IGEN Assay

Interferon (α) and Tumor Necrosis Factor (α) Analysis by ELISA

Interferon (α) concentration is determined by ELISA using a Human Multi-Species kit from PBL Biomedical Laboratories, New Brunswick, NJ. Results are expressed in pg/mL.

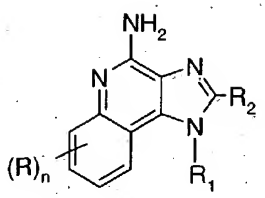
Tumor necrosis factor (α) (TNF) concentration is determined using ELISA kits available from Biosource International, Camarillo, CA. Alternately, the TNF concentration can be determined by ORIGEN M-Series Immunoassay and read on an IGEN M-8 analyzer from IGEN International, Gaithersburg, MD. The

immunoassay uses a human TNF capture and detection antibody pair from Biosource International, Camarillo, CA. Results are expressed in pg/mL.

The complete disclosures of the patents, patent documents, and publications cited herein are incorporated by reference in their entirety as if each were individually incorporated. Various modifications and alterations to this invention will become apparent to those skilled in the art without departing from the scope and spirit of this invention. It should be understood that this invention is not intended to be unduly limited by the illustrative embodiments and examples set forth herein and that such examples and embodiments are presented by way of example only with the scope of the invention intended to be limited only by the claims set forth herein as follows.

WHAT IS CLAIMED IS:

1. A compound of the formula (I):



I

wherein:

R_1 is -X-Q-R_1';

X is -CH(R_4)alkylene or -CH(R_4)alkenylene;

Q is -O-NH-Z-;

Z is selected from the group consisting of:

a bond;

-C(O)-;

-C(S)-;

-$S(O)_2$-;

-$S(O)_2$-N(R_5)-;

-C(O)-O-;

-C(O)-N(R_5)-;

-C(S)-N(R_5)-;

-C(O)-N(R_5)-$S(O)_2$-;

-C(O)-N(R_5)-C(O)-;

-C(S)-N(R_5)-C(O)-; and

-C(O)-C(O)-O-;

R_1' is selected from the group consisting of:

-hydrogen;

-alkyl;

-alkenyl;

-aryl;

-alkylene-aryl;

-alkylene-heteroaryl;

-alkylene-heterocyclyl;

-heteroaryl;

-heterocyclyl; and

alkyl, alkenyl, aryl, arylalkylenyl, heteroarylalkylenyl, heterocyclylalkylenyl, heteroaryl or heterocyclyl, substituted by one or more substituents selected from the group consisting of:

-OH;

-alkyl;

-haloalkyl;

-hydroxyalkyl;

-O-alkyl;

-$S(O)_{0-2}$-alkyl;

-$S(O)_{0-2}$-aryl;

-O-haloalkyl;

-halogen;

-nitrile;

-nitro;

-aryl;

-heteroaryl;

-heterocyclyl;

-O-aryl;

-O-alkylene-aryl;

-C(O)-O-alkyl;

-$C(O)$-$N(R_5)_2$;

-$N(R_5)$-C(O)-alkyl;

-O-(CO)-alkyl; and

-C(O)-alkyl;

n is 0-4;

each R and R_2 are independently selected from the group consisting of hydrogen and non-interfering substituents;

R_4 is selected from the group consisting of hydrogen and alkyl which may be optionally interrupted by one or more -O- groups; and

each R_5 is independently selected from the group consisting of hydrogen, C_{1-10}alkyl, and C_{2-10}alkenyl;

or a pharmaceutically acceptable salt thereof.

2. The compound or salt of claim 1 which induces the synthesis of at least one cytokine.

3. The compound or salt of claim 1 wherein Z is -C(O)-, $-S(O)_2-$, or $-C(O)-N(R_5)-$.

4. The compound or salt of claim 3 wherein R_1' is selected from the group consisting of -alkyl, -alkenyl, -aryl, and heteroaryl, each of which is optionally substituted by one or more substituents selected from the group consisting of -O-alkyl, -S-alkyl, -S-aryl, halogen, -O-C(O)-alkyl, -C(O)-O-alkyl, -haloalkoxy, -haloalkyl, and -aryl.

5. The compound or salt of claim 1 wherein X is $-CH(R_4)C_{1-10}$alkylene.

6. The compound or salt of claim 5 wherein X is propylene or butylene.

7. The compound or salt of claim 1 wherein R_1' is selected from the group consisting of -alkyl, -alkenyl, -aryl, and heteroaryl, each of which is optionally substituted by one or more substituents selected from the group consisting of -O-alkyl, -O-aryl, -S-alkyl, -S-aryl, halogen, -O-C(O)-alkyl, -C(O)-O-alkyl, -haloalkoxy, -haloalkyl, and -aryl.

8. The compound or salt of claim 7 wherein R_1' is selected from the group consisting of -alkyl and -aryl, each of which is optionally substituted by one or more substituents selected from the group consisting of -O-alkyl, -O-aryl, -S-alkyl, -S-aryl, halogen, -O-C(O)-alkyl, -C(O)-O-alkyl; -haloalkoxy, -haloalkyl, and aryl.

9. The compound or salt of claim 1 wherein each R is independently selected from the group consisting of alkyl, alkoxy, halogen, hydroxy, and trifluoromethyl.

10. The compound or salt of claim 1 wherein R_2 is selected from the group consisting of:

-hydrogen;

-alkyl;

-alkenyl;

-aryl;

-heteroaryl;

-heterocyclyl;

-alkylene-Y-alkyl;

-alkylene-Y- alkenyl;

-alkylene-Y-aryl; and

- alkyl or alkenyl substituted by one or more substituents selected from the group consisting of:

-OH;

-halogen;

$-N(R_5)_2$;

$-C(O)-C_{1-10}$alkyl;

$-C(O)-O-C_{1-10}$alkyl;

$-N_3$;

-aryl;

-heteroaryl;

-heterocyclyl;

-C(O)-aryl; and

-C(O)-heteroaryl;

wherein:

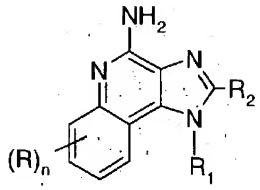
Y is –O– or –$S(O)_{0-2}$-; and

each R_5 is independently selected from the group consisting of hydrogen, C_{1-10}alkyl, and C_{2-10}alkenyl.

11. The compound or salt of claim 10 wherein R_2 is selected from the group consisting of hydrogen, alkyl, and -alkylene-O-alkyl.

12. The compound or salt of claim 1 wherein n is 0.

13. A compound of the formula (I):



I

wherein:

R_1 is -X-Q-R_1';

X is -CH(R_4)alkylene or -CH(R_4)alkenylene;

Q is -O-NH-Z-;

Z is selected from the group consisting of:

a bond;

-C(O)-;

-C(S)-;

-$S(O)_2$-;

-$S(O)_2$-N(R_5)-;

-C(O)-O-;

-C(O)-N(R_5)-;

-C(S)-N(R_5)-;

-C(O)-N(R_5)-S(O)$_2$-;

-C(O)-N(R_5)-C(O)-;

-C(S)-N(R_5)-C(O)-; and

-C(O)-C(O)-O-;

R_1' is selected from the group consisting of:

-hydrogen;

-alkyl;

-alkenyl;

-aryl;

-alkylene-aryl;

-alkylene-heteroaryl;

-alkylene-heterocyclyl;

-heteroaryl;

-heterocyclyl; and

alkyl, alkenyl, aryl, arylalkylenyl, heteroarylalkylenyl, heterocyclylalkylenyl, heteroaryl or heterocyclyl, substituted by one or more substituents selected from the group consisting of:

-OH;

-alkyl;

-haloalkyl;

-hydroxyalkyl;

-O-alkyl;

-S(O)$_{0-2}$-alkyl;

-S(O)$_{0-2}$-aryl;

-O-haloalkyl;

-halogen;

-nitrile;

-nitro;

-aryl;

-heteroaryl;

-heterocyclyl;

-O-aryl;

-O-alkylene-aryl;

-C(O)-O-alkyl;

-C(O)-N(R_5)$_2$;

-N(R_5)-C(O)-alkyl;

-O-C(O)-alkyl; and

-C(O)-alkyl;

n is 0-4;

each R is independently selected from the group consisting of alkyl, alkoxy, halogen, hydroxy, and trifluoromethyl;

R_2 is selected from the group consisting of:

-hydrogen;

-alkyl;

-alkenyl;

-aryl;

-heteroaryl;

-heterocyclyl;

-alkylene-Y-alkyl;

-alkylene-Y- alkenyl;

-alkylene-Y-aryl; and

- alkyl or alkenyl substituted by one or more substituents selected from the group consisting of:

-OH;

-halogen;

-N(R_5)$_2$;

-C(O)-C_{1-10}alkyl;

-C(O)-O-C_{1-10}alkyl;

-N_3;

-aryl;

-heteroaryl;

-heterocyclyl;

-C(O)-aryl; and

-C(O)-heteroaryl;

wherein:

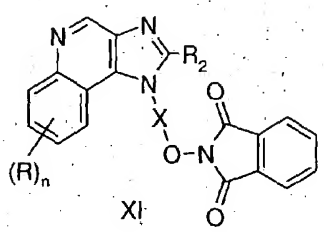
Y is –O– or –$S(O)_{0-2}$-;

R_4 is selected from the group consisting of hydrogen and alkyl which may be optionally interrupted by one or more -O- groups; and

each R_5 is independently selected from the group consisting of hydrogen, C_{1-10}alkyl, and C_{2-10}alkenyl;

or a pharmaceutically acceptable salt thereof.

14. A compound of the formula (XI):



XI

wherein:

X is -CH(R_4)alkylene or -CH(R_4)alkenylene;

n is 0-4;

each R is independently selected from the group consisting of alkyl, alkoxy, halogen, hydroxy, and trifluoromethyl; and

R_2 is selected from the group consisting of:

-hydrogen;

-alkyl;

-alkenyl;

-aryl;

-heteroaryl;

-heterocyclyl;

-alkylene-Y-alkyl;

-alkylene-Y- alkenyl;

-alkylene-Y-aryl; and

- alkyl or alkenyl substituted by one or more substituents selected from the group consisting of:

-OH;

-halogen;

$-N(R_5)_2$;

$-C(O)-C_{1-10}$alkyl;

$-C(O)-O-C_{1-10}$alkyl;

$-N_3$;

-aryl;

-heteroaryl;

-heterocyclyl;

-C(O)-aryl; and

-C(O)-heteroaryl;

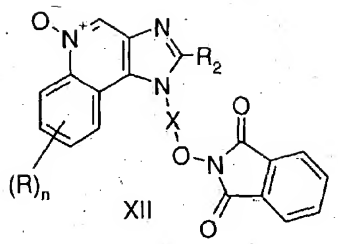
wherein:

Y is –O– or $-S(O)_{0-2}-$;

R_4 is selected from the group consisting of hydrogen and alkyl which may be optionally interrupted by one or more -O- groups; and

each R_5 is independently selected from the group consisting of hydrogen, C_{1-10}alkyl, and C_{2-10}alkenyl.

15. A compound of the formula (XII):



XII

wherein:

X is -CH(R_4)alkylene or -CH(R_4)alkenylene;

n is 0-4;

each R is independently selected from the group consisting of alkyl, alkoxy, halogen, hydroxy, and trifluoromethyl; and

R_2 is selected from the group consisting of:

-hydrogen;

-alkyl;

-alkenyl;

-aryl;

-heteroaryl;

-heterocyclyl;

-alkylene-Y-alkyl;

-alkylene-Y- alkenyl;

-alkylene-Y-aryl; and

- alkyl or alkenyl substituted by one or more substituents selected from the group consisting of:

-OH;

-halogen;

-$N(R_5)_2$;

-C(O)-$C_{1\text{-}10}$alkyl;

-C(O)-O-$C_{1\text{-}10}$alkyl;

-N_3;

-aryl;

-heteroaryl;

-heterocyclyl;

-C(O)-aryl; and

-C(O)-heteroaryl;

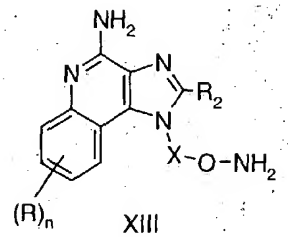
wherein:

Y is –O– or –$S(O)_{0-2}$-;

R_4 is selected from the group consisting of hydrogen and alkyl which may be optionally interrupted by one or more -O- groups; and

each R_5 is independently selected from the group consisting of hydrogen, C_{1-10}alkyl, and C_{2-10}alkenyl.

16. A compound of the formula (XIII):



XIII

wherein:

X is -$CH(R_4)$alkylene or -$CH(R_4)$alkenylene;

n is 0-4;

each R is independently selected from the group consisting of alkyl, alkoxy, halogen, hydroxy, and trifluoromethyl; and

R_2 is selected from the group consisting of:

-hydrogen;

-alkyl;

-alkenyl;

-aryl;

-heteroaryl;

-heterocyclyl;

-alkylene-Y-alkyl;

-alkylene-Y- alkenyl;

-alkylene-Y-aryl; and

- alkyl or alkenyl substituted by one or more substituents selected from the group consisting of:

-OH;

-halogen;

-$N(R_5)_2$;

-$C(O)$-$C_{1\text{-}10}$alkyl;

-$C(O)$-O-$C_{1\text{-}10}$alkyl;

-N_3;

-aryl;

-heteroaryl;

-heterocyclyl;

-C(O)-aryl; and

-C(O)-heteroaryl;

wherein:

Y is –O– or –$S(O)_{0\text{-}2}$-;

R_4 is selected from the group consisting of hydrogen and alkyl which may be optionally interrupted by one or more -O- groups; and

each R_5 is independently selected from the group consisting of hydrogen, $C_{1\text{-}10}$alkyl, and $C_{2\text{-}10}$alkenyl.

17. A pharmaceutical composition comprising a therapeutically effective amount of a compound of claim 1 in combination with a pharmaceutically acceptable carrier.

18. A pharmaceutical composition comprising a therapeutically effective amount of a compound of claim 13 in combination with a pharmaceutically acceptable carrier.

19. A method of inducing cytokine biosynthesis in an animal comprising administering an effective amount of a compound of claim 1 to the animal.

20. A method of inducing cytokine biosynthesis in an animal comprising administering an effective amount of a compound of claim 13 to the animal.

21. A method of treating a viral disease in an animal in need thereof comprising administering a therapeutically effective amount of a compound of claim 1 to the animal.

22. A method of treating a viral disease in an animal in need thereof comprising administering a therapeutically effective amount of a compound of claim 13 to the animal.

23. A method of treating a neoplastic disease in an animal in need thereof comprising administering a therapeutically effective amount of a compound of claim 1 to the animal.

24. A method of treating a neoplastic disease in an animal in need thereof comprising administering a therapeutically effective amount of a compound of claim 13 to the animal.

HYDROXYLAMINE SUBSTITUTED IMIDAZOQUINOLINES

ABSTRACT OF THE DISCLOSURE

Imidazoquinoline compounds with a hydroxylamine substituent at the 1-position, pharmaceutical compositions containing the compounds, intermediates, and methods of use of these compounds as immunomodulators, for inducing cytokine biosynthesis in animals and in the treatment of diseases including viral and neoplastic diseases are disclosed.